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=> e hohn b/au

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E12 97 HOHN D C/AU

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=> s e3

L1 205 "HOHN B"/AU

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E1 1 HINNELBUSCH B J/AU
E2 1 HINNELLS P J G/AU
E3 72 --> HINNEN A/AU
E4 2 HINNEN BOUWMANS C/AU
E5 7 HINNEN D/AU
E6 2 HINNEN D A/AU
E7 13 HINNEN F/AU
E8 1 HINNEN H/AU
E9 4 HINNEN M/AU
E10 2 HINNEN M G J/AU
E11 13 HINNEN M L/AU
E12 16 HINNEN R/AU

=> s e3

L2 72 "HINNEN A"/AU

=> s l1 and l2

L3 0 L1 AND L2

=> s l1 and yeast and cosmid

L4 0 L1 AND YEAST AND COSMID

=> s l2 and yeast and cosmid

L5 0 L2 AND YEAST AND COSMID

=> s l1 and yeast and vector

L6 0 L1 AND YEAST AND VECTOR

=> s l2 and yeast and vector

L7 6 L2 AND YEAST AND VECTOR

=> d 1-6 bib ab

L7 ANSWER 1 OF 6 MEDLINE

AN 91029620 MEDLINE

DN 91029620

TI The influence of GAP promoter variants on hirudin production,
average plasmid copy number and cell growth in *Saccharomyces*
cerevisiae.

AU Janes M; Meyhack B; Zimmermann W; Hinnen A

CS Ciba-Geigy AG, Biotechnology Department, Basel, Switzerland..
 SO CURRENT GENETICS, (1990 Aug) 18 (2) 97-103.
 Journal code: CUG. ISSN: 0172-8083.
 CY United States
 DT Journal; Article; (JOURNAL ARTICLE)
 LA English
 FS Priority Journals
 EM 199102
 AB The **yeast** *Saccharomyces cerevisiae* has been engineered to synthesize and secrete desulfato-hirudin (hirudin), a thrombin inhibitor from the leech *Hirudo medicinalis*. The synthetic gene coding for hirudin was expressed constitutively under the control of four size-variants of the **yeast** glyceraldehyde-3-phosphate dehydrogenase promoter (GAP) and cloned into a 2 mu based multicopy **yeast vector**. The constitutive action of the four promoter variants was confirmed by demonstrating that the expression and secretion of hirudin is growth-related. The different efficiencies of the promoter variants not only affected hirudin expression but also led to changes in several cellular parameters, such as cell growth, average plasmid copy number and plasmid stability. The observed changes show that **yeast** cells establish a specific equilibrium for each promoter variant. We conclude, that the adjustment of cellular parameters in response to the expression levels of a heterologous protein is regulated by two counteracting selective forces: (1) the need for complementation of the auxotrophic host marker by the plasmid-encoded selection gene which, in the case of dLEU2, requires several plasmid copies; and (2) a selective advantage of cells with a lower copy number enabling them to escape the burden of heterologous protein production.

L7 ANSWER 2 OF 6 MEDLINE
 AN 90028909 MEDLINE
 DN 90028909
 TI Heterologous gene expression in **yeast**.
 AU **Hinnen A**; Meyhack B; Heim J
 SO BIOTECHNOLOGY, (1989) 13 193-213. Ref: 63
 Journal code: BIT. ISSN: 0740-7378.
 CY United States
 DT Journal; Article; (JOURNAL ARTICLE)
 General Review; (REVIEW)
 (REVIEW, TUTORIAL)
 LA English
 FS Priority Journals
 EM 199002

L7 ANSWER 3 OF 6 MEDLINE
 AN 89289703 MEDLINE
 DN 89289703
 TI Functional analysis of the signal-sequence processing site of **yeast** acid phosphatase.
 AU Monod M; Haguenauer-Tsapis R; Rauseo-Koenig I; **Hinnen A**
 CS Service de Dermatologie, CHUV, Lausanne.
 SO EUROPEAN JOURNAL OF BIOCHEMISTRY, (1989 Jun 15) 182 (2) 213-21.
 Journal code: EMZ. ISSN: 0014-2956.
 CY GERMANY, WEST: Germany, Federal Republic of
 DT Journal; Article; (JOURNAL ARTICLE)
 LA English
 FS Priority Journals; Cancer Journals
 EM 198910
 AB A systematic study of the signal peptidase cleavage site of the main

cell-wall-repressible *Saccharomyces cerevisiae* acid phosphatase encoded by the PHO5 gene is presented. The last amino acid of the signal sequence, the chromosomally encoded alanine of the wild-type gene, was changed by any of 19 other amino acids in the chromosomal DNA by using in vitro mutagenesis in *Escherichia coli* and the technique of gene replacement. Processing and secretion are normal when the amino acid at this position is a small neutral amino acid, i.e. alanine, glycine, cysteine, serine or threonine. Processing glycosylation, and secretion of regulated acid phosphatase are distinctly affected with other amino acid substitutions and core-glycosylated protein accumulates in the cell. Surprisingly, PHO5 protein is still secreted to the cell wall and into the growth medium but at a lower rate and without cleavage of the signal sequence. The same features are exhibited by a mutated acid phosphatase with a deletion of four amino acids at the end of the signal peptide (-7 to -4 relative to the processing site) thus preserving the important -3 to -1 region.

L7 ANSWER 4 OF 6 MEDLINE
 AN 82116250 MEDLINE
 DN 82116250
 TI **Vectors** for cloning in **yeast**.
 AU **Hinnen A**; Meyhack B
 SO CURRENT TOPICS IN MICROBIOLOGY AND IMMUNOLOGY, (1982) 96 101-17.
 Ref: 64
 Journal code: DWQ. ISSN: 0070-217X.
 CY GERMANY, WEST:-Germany, Federal Republic of
 DT Journal; Article; (JOURNAL ARTICLE)
 General Review; (REVIEW)
 LA English
 EM 198206

L7 ANSWER 5 OF 6 BIOSIS COPYRIGHT 1998 BIOSIS
 AN 90:494058 BIOSIS
 DN BA90:122404
 TI THE INFLUENCE OF GAP PROMOTER VARIANTS ON HIRUDIN PRODUCTION AVERAGE PLASMID COPY NUMBER AND CELL GROWTH IN *SACCHAROMYCES-CEREVISIAE*.
 AU JAMES M; MEYHACK B; ZIMMERMANN W; **HINNEN A**
 CS CIBA-GEIGY AG, BIOTECHNOL. DEP., K-681.1.44, P.O. BOX, CH-4002 BASEL, SWITZERLAND.
 SO CURR GENET 18 (2). 1990. 97-104. CODEN: CUGED5 ISSN: 0172-8083
 LA English
 AB The **yeast** *Saccharomyces cerevisiae* has been engineered to synthesize and secrete desulfato-hirudin (hirudin), a thrombin inhibitor from the leech *Hirudo medicinalis*. The synthetic gene coding for hirudin was expressed constitutively under the control of four size-variants of the **yeast** glyceraldehyde-3-phosphate dehydrogenase promoter (GAP) and cloned into a 2 .mu. based multicopy **yeast vector**. The constitutive action of the four promoter variants was confirmed by demonstrating that the expression and secretion of hirudin is growth-related. The different efficiencies of the promoter variants not only affected hirudin expression but also led to changes in several cellular parameters, such as cell growth, average plasmid copy number and plasmid stability. The observed changes show that **yeast** cells establish a specific equilibrium for each promoter variant. We conclude, that the adjustment of cellular parameters in response to the expression levels of a heterologous protein is regulated by two counteracting selective forces: (1) the need for complementation of the auxotrophic host marker by the plasmid-encoded selection gene

which, in the case of dLEU2, requires several plasmid copies; and (2) a selective advantage of cells with a lower copy number enabling them to escape the burden of heterologous protein production.

L7 ANSWER 6 OF 6 BIOSIS COPYRIGHT 1998 BIOSIS
AN 82:130472 BIOSIS
DN BR23:60464
TI **VECTORS** FOR CLONING IN **YEAST** SACCHAROMYCES-
CEREVISIAE.
AU **HINNEN A**; MEYHACK B
CS FRIEDRICH MIESCHER-INST., P. O. BOX 273, CH-4002 BASEL, SWITZERLAND.
SO HOFSCHEIDER, P. H. AND W. GOEBEL (ED.). CURRENT TOPICS IN
MICROBIOLOGY AND IMMUNOLOGY, VOL. 96. GENE CLONING IN ORGANISMS OTHER
THAN E.COLI. VII+259P. SPRINGER-VERLAG: BERLIN, WEST GERMANY; NEW
YORK, N.Y., USA. ILLUS. 0 (0). 1982. P101-118. CODEN: CTMIA3 ISBN:
3-540-11117-4; 0-387-11117-4 ISSN: 0070-217X
LA English